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## **Can oral infection be a risk factor for Alzheimer's disease?**

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## ABSTRACT

Alzheimer's disease (AD) is a **scourge** of longevity that will drain enormous resources from public health budgets in the future. Currently, there is no diagnostic biomarker and/or treatment for this most common form of dementia in **humans**. AD can be of early familial-onset or sporadic with a late-onset. Apart from the two main hallmarks, amyloid-beta and ~~the~~ neurofibrillary tangles, inflammation is a characteristic feature of AD neuropathology. Inflammation may be caused by a local central nervous system insult and/or by peripheral infections. Numerous **microorganisms** are suspected in AD brains ranging from bacteria (mainly oral and non-oral *Treponema* species), viruses (Herpes simplex type I) and yeasts (*Candida* species). A causal relationship between periodontal pathogens/non-oral *Treponema* species of bacteria has been proposed via the amyloid-beta and inflammatory links. Periodontitis constitutes a peripheral oral infection that can provide the brain with intact bacteria and virulence factors and inflammatory mediators due to daily, transient bacteraemias. If and when genetic risk factors meet environmental risk factors in the brain, disease is expressed, in which neurocognition may be ~~exacerbated~~ impacted, leading to the of dementia. To achieve the goal of finding a diagnostic biomarker and possible prophylactic treatment for AD, there is an initial need to solve the etiological puzzle contributing to its pathogenesis. This review therefore addresses oral infection as the plausible aetiology of late onset AD (LOAD). ~~the plausible aetiology of the late-onset AD being an oral infection.~~

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Keywords: Alzheimer's disease; pathogenesis; microorganisms; oral bacteria; direct cause

Alzheimer's disease (AD) is a neurodegenerative disease and the most common example of a group of diseases that manifest as dementia. It is associated with atrophy and specific neuronal death particularly in the hippocampal region of the brain (1). Research into AD pathogenesis, has flagged two main categories of the disease: ~~A;~~ the familial-onset-onset ~~that~~ accounts for around 2% of all AD cases and the sporadic form of late-onset AD also to as LOAD ~~that~~ constitutes approximately 98% of the cases. LOAD displays genetic susceptibility traits of which the well-known risk factor is inheritance of the apolipoprotein (*APOEε4*) gene allele (2) and, appears to require an environmental factor for disease expression. For example a pathogen-host interaction, can exacerbate neurocognition in some elderly individuals who if in their 80+ years likely become diagnosed with LOAD (3, 4). The rationale for this review therefore is to try to explain the aetiology in the vast proportion of LOAD cases that relies upon common risk factors, ~~and to date, several~~ Several scientists these to be peripheral infections (5-11); and the accompanying systemic and local inflammatory mediators (11-13). Of these, the plausible risk from oral infection is the main focus of this review.

57

## 58 PREVALENCE OF AD

AD is a seource-burden of longevity resulting from the superior quality of health care This factor is likely to contribute to quadrupling of AD subjects living in our society during the next 40 years (14). It is estimated that by 2050 about 13-14 million people are likely to suffer from AD in the USA with a rise in the total costs estimated to be more than \$1 trillion. The odds of having a diagnosis of AD when over 85 years of age exceed 1:3 (15). One in six people over 80 years in the UK ~~have-has~~ dementia (16). Estimates for the prevalence of AD in USA indicate that more than 5 million individuals who are 65 years or older currently suffer from AD (1, 15). About 200,000 subjects have been diagnosed with the early-onset familial

AD form and health care costs for this disease are about \$200 billion per year (1). It is clear that AD is fast becoming a major health challenge in the USA and around the globe that will financially drain public health budgets and care giver services.

## NEUROPATHOLOGICAL CHARACTERISTICS OF THE AD BRAIN

The AD brain is characterized by several neuropathological features of which two seminal hallmarks (Fig. 1) arise from proteostasis of the ongoing neurodegenerative processes and are essential for a definitive diagnosis of the disease ~~at~~ post mortem (17). One of the hallmark proteins is made up of fibrils in the form of extracellular, insoluble plaques and consists primarily of amyloid-beta ( $A\beta$ ) (18). The se peptide deposits in variable sizes depend upon the secretase enzymes ( $\alpha$ -,  $\beta$ - and  $\gamma$ -secretases) that cleave it from the longer amyloid precursor protein (APP). Initial reports suggested fibrillar  $A\beta$  to be neurotoxic (19) as it has been shown to kill all types of cells by apoptosis induction (20). However, there are two known insoluble fibrillar  $A\beta$  amyloid peptides comprised of  $A\beta_{40}$  and  $A\beta_{42}$  amino-acid residues as well as their different which exhibit distinct physiological states within the human brain. There is a general consensus among scientists that the larger ( $A\beta_{42}$ ) peptide is the neurotoxic form as the ageing brain of cognitive intact individuals also displays  $A\beta$  plaques. However, in the cognitively intact brain they are fewer in number and usually of the diffuse  $A\beta_{40}$  type that appears not to bear any, as yet known, pathological significance ~~in the elderly who age successfully.~~ In monomeric, dimeric and the multimeric forms of  $A\beta$  (21). The relative neurotoxicity of these isoforms remains unclear ~~It is not clear as to which one of these is more neurotoxic~~ (22).

More recently, the fibrillary forms of the  $A\beta_{(40/42)}$  peptides released in the AD brain ~~are~~ were also recognized as “defensin” or innate immune defense molecules that act to protect the host against infection (23). For example, both of the aforementioned amyloidogenic peptides can

bind to bacterial membranes and in that way lyse bacterial cells. Although A $\beta$  is acting as an antimicrobial peptide (AMP), it may be a part of the brain's ancient/modern innate immune defense mechanism. AMPs are potent, broad-spectrum, pore-forming agents ~~against~~ targeting Gram-negative and Gram-positive bacteria, enveloped viruses and protozoans (23), thereby supporting the hypothesis that AD has an infectious origin.

Furthermore, the senile plaques (A $\beta$ <sub>42</sub>) are recognized as triggers that stimulate activation of microglial cells and initiate local immune responses (24). Activated microglia are the most important contributors of inflammation in the central nervous system (CNS) (25). They secrete a number of proinflammatory cytokines (24-26) and recognize pattern associated molecular patterns (PAMPs) on bacteria and their cellular debris (27-30) ~~to deal with in~~ response to CNS infection.

The other pathological characteristic of AD is an accumulation of intracellular hyperphosphorylated tau and heat shock proteins constituting the neurofibrillary tangles (NFTs). Hyperphosphorylated tau protein alters the polymerization and stability of microtubules compromising their function (31). NFTs in AD reflect the severity of disease; however, the significance of pathogen-host interaction to the occurrence of NFTs in the AD brain is poorly understood. Current genetic evidence is pointing to aberrant innate immune responses (32, 33) and cholesterol lipid genes (~~see~~ 34) having greater significance in AD pathogenesis. A dysfunctional immune system and predisposition to hyperlipidaemia also support the role of reduced blood flow due to the vascular lesions and inflammation, A $\beta$  deposition and microorganisms in AD.

In advanced AD pathology, synaptic dysfunction is another structural defect associated with a decline in memory (35-37). Although a circular argument, malnutrition plays a role in the gradual loss of synapses and fewer teeth during life is a known risk factor for AD (38).

Neurons are capable of responding to injury by expressing multiple neurotransmitters. In AD, selective loss of cholinergic neurons in the basal forebrain (39) also correlates with the loss of cognitive function (18, 35).

## THE AMYLOID CASCADE HYPOTHESIS

Several hypotheses have been ~~launched for~~ advanced regarding the development of AD. The amyloid cascade hypothesis serves as a model particularly for the familial form of AD (40) which is a disease caused by mutations involving the amyloid- $\beta$  protein precursor, located on chromosome 21 and presenilin 1 and 2 on chromosomes 14 and 1 respectively that enhance the APP gene processing towards A $\beta$  deposition (41, 42). The model, which was first proposed by Glenner and Wong (43), maintains that the neurodegenerative disease is due to an imbalance between the generation and clearance of A $\beta$ . Genome wide association studies (GWAS) highlighted the complement receptor 1 (*CR1*) gene playing a role in AD pathogenesis (44). One recognized role of CR1, a membrane bound regulatory protein, is its ability to bind C3b opsonins (Fig. 2). It is abundantly expressed especially on erythrocyte membranes and as such participates in immune complex clearance by transporting waste to the liver and the spleen. As the *CR1* gene is a risk factor for LOAD, this suggests loss of function as a possibility for the defective clearance of A $\beta$  in the brain. Other tentative explanations suggest variation in CR1 protein isoforms (longer and shorter forms) (45), whereby the longer form is somehow negatively less involved in the disease process via its ability to bind more C3b and facilitate more effective clearance of A $\beta$  in the brain (46). This is a process that inevitably fails favoring disease expression with more A $\beta$  proteostasis buildup and complement pathway activation. The amyloid hypothesis has been modified several times, particularly due to the finding that soluble oligomers of A $\beta$  may contribute to

early preclinical stages of the disease that initiate the cascade leading to synaptic dysfunction, atrophy and neuronal loss (47).

## THE INFLAMMATORY HYPOTHESIS

### The intrinsic model

Currently there are two models of the inflammatory hypothesis of AD, an intrinsic and an extrinsic. The intrinsic inflammation model accounts for the intact “blood-brain barrier” (BBB) restricting entry of neurotoxic immune molecules and systemic lymphocytes to the brain. As a consequence, the brain glial cells are able to generate a local and complete innate immune system when challenged by foreign agents (26, 48-50). Historically, neuroinflammation has largely been viewed as being a downstream consequence of the amyloid hypothesis, whereby the presence of amyloidogenic peptides result in the activation of microglia initiating pro-inflammatory cascades and the release of potentially neurotoxic substances resulting in degenerative changes in neurons. GWAS now implicates innate immune genes (44, 51) as being a risk factor and supports a primary role for the inflammatory elements of AD pathology via inappropriate activation of the complement system (52-54) in association with A $\beta$  plaques and NFTs (55).

### The extrinsic model

The extrinsic model accounts for communication of the glial cells with the immune challenges presented via the blood vascular system using the circumventricular organs and the choroid plexus that are devoid of the BBB (56). The cells from this region of the brain are fully equipped with the CD14 receptor and the toll-like receptor 4 (TLR 4) to recognise recognize the peripheral blood circulation (27, 28). Hence, elements of systemic infections such as those



164 originating from Gram-negative, highly virulent oral pathogens, bronchopneumonia and  
 165 urinary tract infections (3, 4, 7, 57, 58) reach all organs including the CNS. ~~The consequences~~  
 166 products entering the bloodstream ~~trigger the~~ ~~are that the~~ innate immune responses of ~~hosts~~  
 167 ~~pattern recognition receptors (PPR) and TLRs via pattern recognition receptors (PPR) and~~  
 168 ~~infectious threat by secreting~~ ~~to the threat of infection by secreting~~ immune mediators  
 169 agents. Increased risk of dementia in the elderly following multiple infectious episodes has  
 170 been reported ~~It is reported that multiple episodes of infections in the elderly likely end up~~  
 171 ~~being diagnosed with dementia~~ (4). In addition, systemic infections appear to contribute  
 172 towards delirium in some clinically diagnosed AD patients and such episodes can exacerbate  
 173 a premorbid cognitive status (3). Holmes et al. proposed that since cytokines are primary  
 174 mediators released by the host to defend against infection, such secondary stimuli (IL-1 $\beta$  and  
 175 TNF- $\alpha$ ) may mediate their effect on the brain and indirectly contribute to cognitive decline (3,  
 176 57).

Commented [PB1]: Break up to 2 sentences.

177

## 178 NON-ORAL BACTERIA RELATED TO AD

179 Honjo et al. (59) using Bradford Hill's criteria for assessing the relationship between bacteria  
 180 and disease found *Chlamydomphila pneumoniae* to be a likely infectious agent related to the  
 181 pathogenesis of AD. Maheshwari and Eslick (60) reported a strong correlation between *C.*  
 182 *pneumoniae* and AD, and according to Shima et al. (61) *C. pneumoniae* is currently the most  
 183 plausible of all infectious agents proposed to be involved in AD. Lim et al. (62) suggested that  
 184 the pro- and chronic inflammatory states in AD pathogenesis may in part be due to *C.*  
 185 *pneumoniae* infection of monocytes. *C. pneumoniae* antibodies from typical intracellular and  
 186 atypical *C. pneumoniae* antigens have been identified both ~~from typical intracellular and~~  
 187 of ~~the brains~~ ~~from~~ AD patients (63). Amyloid deposit and NFTs were detected in the same

188 regions in apposition to one another suggesting that *C. pneumoniae* infection is involved in  
 189 the development of AD pathology.

190 Using various techniques Balin et al. (9) found *C. pneumoniae* in 80-90% of LOAD brain  
 191 tissue specimens. *C. pneumoniae* infection was correlated with the *APOEε4* allele expression.  
 192 The same researchers subsequently demonstrated that astroglia, microglia, neurons,  
 193 endothelial cells and monocytes in the LOAD brain are permissive to this bacterium. The  
 194 mechanisms of pathogenesis differ between actively- and persistently-infecting chlamydiae  
 195 and it is in the persistent state that these organisms cause chronic disease (64, 65). *C.*  
 196 *pneumoniae* was cultured from two AD brain samples after one or two passages in HEp-2  
 197 cells (66). Interestingly, the study indicated that brain isolates were more related to respiratory  
 198 than to vascular/atheroma strains of *C. pneumoniae*. This suggested that *C. pneumoniae*  
 199 infection of the brain was secondary to bronchopneumonia and at the end stages of LOAD.

200 It has been suggested that the phages phiCPAR39 and phiCPG1, associated with *C.*  
 201 *pneumoniae*, may enter mitochondria of the bacterial host and work as slow viruses initiating  
 202 AD (67). These authors hypothesized that mitochondrial recruitment by *C. pneumoniae*  
 203 phages may be the primary initiating event in the pathogenesis of neurodegenerative  
 204 disorders.

205 In a meta-analysis based on 25 relevant, primarily case-control studies Maheshwari and Eslick  
 206 (60) found a statistically significant association between AD and detectable evidence of  
 207 infection caused by *C. pneumoniae* or spirochetes. They reported over a ten-fold increased  
 208 occurrence of AD when there was evidence of spirochetal infection (OR: 10.61; 95% CI:  
 209 3.38-33.29) and over a four-fold increased occurrence of AD with a conservative risk estimate  
 210 (OR: 4.45; 95% CI: 2.33-8.52). There was a five-fold increase in occurrence of AD with *C.*  
 211 *pneumoniae* infection (OR: 5.66; 95% CI: 1.83-17.51). Accordingly, a strongly positive

association between bacterial infection and AD was shown for both types of bacteria, but it was strongest for spirochetes.

It is generally accepted that the syphilis spirochete *Treponema pallidum* can cause chronic neuropsychiatric disorders including dementia as well as other neurodegenerative disorders (11). *T. pallidum* causes brain atrophy and A $\beta$  deposition in the atrophic form of general paresis (68, 69) and is a strong indication for involvement of spirochetes in AD pathogenesis. Chronic diseases such as syphilis are frequently associated with deposition of amyloid (68, 69). ~~Actually, amyloid is considered as~~ Amyloid is an integral ~~part~~ component of spirochetes which may contribute to amyloid deposition in AD (70). ~~Syphilis accumulation of spirochetes~~ Spirochete accumulation in the cerebral cortex ~~in the context of syphilis~~ will also lead to formation of senile plaques, NFTs and granulovacuolar degeneration (71).

Miklossy (68, 69) analyzed data on the ability of spirochetes to induce pathological and biological hallmarks of AD *in vitro* following Koch's and Hill's postulates and demonstrated a plausible causal relationship between neurospirochetosis and AD. The data revealed a statistically significant association between spirochetes and AD ( $P = 1.5 \times 10^{-17}$ , OR = 20, 95% CI = 8-60, N = 247). When mammalian cells were exposed to spirochetes, the pathological and biological hallmarks of AD were reproduced *in vitro* (68, 69). ~~Miklossy (72) also found that historical~~ Historical observations supported the conclusion ~~that—that~~ observations paved the way for drawing conclusions such as chronic spirochetal infections can cause dementia and reproduce the neuropathological hallmarks of AD (72). According to Miklossy (72), these observations represent further evidence in support of a causal relationship between various spirochetal infections and AD.

Another spirochete ~~also implicated in AD is~~ *Borrelia burgdorferi*, ~~has also been implicated in~~ AD. This is the causative agent of Lyme disease, ~~which is~~ which is transfected to humans via

tick vectors ~~through infected tick bites~~. There are great similarities in the clinical and syphilis and Lyme disease (72, 73). The occurrence of *B. burgdorferi* in the brains of AD patients was first reported by MacDonald and Miranda (74) and was confirmed later by MacDonald (75, 76), Riviere et al. (5) and Miklosy et al. (77). Interestingly, Bu et al. (78) found that the infectious burden consisting of *B. burgdorferi*, *C. pneumoniae*, *Helicobacter pylori*, ~~cytomegalovirus~~ ~~cytomegalo virus~~ and Herpes simplex-1 (HSV-1) is associated with Gutacker et al. (79) and Pappolla et al. (80) found no evidence for an association between *B. burgdorferi* and AD.

Among other bacterial species, *H. pylori* ~~alone~~ (monoinfection) has been found to be related to AD (59). These authors suggested that AD pathology can be initiated and exacerbated by some microorganisms with inflammatory and oxidative responses which may affect the brain continuously and gradually over time. However, the *H. pylori* status ~~did not depend on~~ was not associated with AD in a study from Japan, probably due to the high prevalence of the organism in controls (81). This was refuted by Kountouras et al. (82) who had previously found that successful eradication of *H. pylori* infection was associated with significantly lower mortality risk in AD patients [HR (95% CI)=0.287 (0.114-0.725), p=0.008] (83).

Commented [PB2]: Unclear. Do you mean as a monoinfection?

## ORAL BACTERIA RELATED TO AD

The oral cavity ~~harbours~~ harbors an impressive range of bacterial phylotypes (84). Molecular identification methods have detected close to 900 different predominant bacterial species of which 35% cannot yet be cultured (85). The oral microbiome profiles appear to be individualized (86), meaning that bacterial microbiomes can vary both qualitatively and quantitatively between individuals, although there are also significant overlaps. Each individual can ~~harbour~~ harbor up to 200 different bacterial taxa in their mouth and there is a

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Commented [PB4]: There are differences however there is also significant overlap.

variation in the microbiota in different oral sites (84, 87). Furthermore, the composition of the oral microbiota irrespective of being indigenous or pathogenic in the oral cavity keeps changing in view of major oral diseases (caries, gingivitis, aggressive and chronic periodontitis, periodontal-endodontic lesions, peri-implantitis and mucositis) (88-94). Particularly plaque-induced oral diseases such as periodontitis are associated with a change in the oral microbiota. There is a predominance of anaerobic bacteria in the oral cavity. Many of the major periodontal microorganisms are anaerobic, e.g., *Porphyromonas gingivalis*, *Treponema denticola* and *Tannerella forsythia*. The abundance of anaerobes tend to increase with the development of plaque-induced oral diseases.

#### Periodontal bacterial pathogens are related to AD

Major pathogens of chronic periodontitis such as *P. gingivalis*, *T. forsythia*, and *T. denticola* are implicated in the development of several inflammatory diseases at remote organ sites. Except for *T. forsythia*, all the above three of the above-named organisms of which *T. denticola* represents a spirochetes, have been found in the AD brain (5, 8). Spirochetes are strongly neurotropic. They can spread along nerve fibers and via lymphatics (67, 68) and have been detected in the trigeminal nerve and trigeminal ganglia (95). Spirochetes and their antigens as well as DNA have been found associated with AD and are strongly implicated as the causative agents leading to dementia (68, 69). In 14 studies spirochetes were detected in AD by different authors in different laboratories and countries by means of different techniques (for a review see Miklossy (68, 69). Riviere et al. (5) demonstrated the presence of seven different oral *Treponema* species in 14 out of 16 AD brain specimens (Fig. 3). Spirochetes were even cultivated from the brains of AD patients indicating that they were viable in the brain (67, 68, 77). Miklossy suggested a co-infection by several spirochetes in

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AD including the oral varieties (*T. socranskii*, *T. pectinovorum*, *T. denticola*, *T. medium*, *T. amylovorum* and *T. maltophilum*) as demonstrated by Riviere et al. (5). Spirochetes reproduced the biological and pathological hallmarks of AD after exposure of mammalian neuronal and glial cells in organotypic cultures (68, 69).

It ~~has been~~ was demonstrated that LPS from periodontal bacteria can access the AD brain during life ~~as while~~ detection in corresponding controls, with equivalent or longer postmortem interval was absent (8). This study supports the literature on elevated antibodies to periodontal disease-associated bacteria such as *P. gingivalis*, being found in AD patients (7). Furthermore, in 2,355 people 60 years and over, the third NHANES study found associations between periodontitis and cognitive impairment and between measures of immunoglobulin to *P. gingivalis* and cognitive test performance (96, 97)-~~used cohort methodology analyzing serum levels of antibodies to periodontal disease. All~~ In this study all participants were cognitively intact at baseline. Those who went on to develop AD had higher levels of serum antibodies to periodontal pathogens at baseline. ~~This~~ The study ~~suggested~~ suggested a temporal periodontal disease came before AD.

Other important periodontal pathogens related to AD are *Fusobacterium nucleatum* and *Prevotella intermedia*. In the NHANES study antibody ~~Antibody~~ levels to these organisms were significantly increased ( $\alpha = 0.05$ ) at baseline serum in patients with AD compared to controls (97). The results were significant after controlling for baseline age, Mini-Mental State Examination score, and allele *APOE* $\epsilon$ 4 status. Noble et al. (98) found that a high anti-*Actinomyces naeslundii* titer (> 640 ng/ml, present in 10% of the subjects) was associated with increased risk of AD (HR=2.0, 95% CI: 1.1-3.8). This association was stronger after adjusting for other significant titers (HR=3.1, 95%CI: 1.5-6.4) and confirmed that periodontal pathogens ~~can~~ may be associated with AD.

### Possible consequences to the brain of carrying oral bacterial pathogens

The fact that inflammation is sustained in the AD brain suggests that local immunogenic hallmark proteins and/or peripheral infections are key perpetrators. This is supported by reports highlighting microorganisms and their toxic products as well as DNA in brain tissue of AD patients and experimental animals (see [later below](#)). Bacteria activate pathways that include the integrin receptor CR3 (CD11b/CD18) and TLR signalling (99) and the complement cascade (100). The NF- $\kappa$ B signalling pathway for cyto/chemokine release (TNF- $\alpha$ , IL-8) (101) produces free radicals, nitric oxide [triggers](#) and apoptosis (102). The oral cavity, lungs and gastrointestinal and urinary tracts are plausible sources of brain microorganisms. The likely passage of the microorganisms of interest from their original sites to the brain is described below.

Infections with spirochetes can cause cerebral hypoperfusion (103), cerebrovascular lesions and a severely disturbed capillary network (68, 69). Chronic spirochetal infections can also induce slowly progressive dementia, cortical atrophy, chronic inflammation and A $\beta$  deposition, ~~which cannot be distinguished~~ [indistinguishable](#) from that occurring in AD brains (for reviews see 68, 69, 72). Furthermore, cultured neuronal cells exposed to spirochetes produce A $\beta$  (104). Spirochetes are also able to form plaque-, tangle- and curly fiber-like lesions (72, 105). [They induce a latent and slowly progressive infection by evading host defenses. This promotes their survival and proliferation in the brain by blocking the complement cascade. Spirochetes may even survive and proliferate in hosts that are immune-competent.](#) ~~By evading host's defenses, spirochetes induce a latent and slowly progressive infection to promote their survival and proliferation in the brain and by blocking the complement cascade spirochetes may survive and proliferate even in hosts that are immune-competent.~~ Interestingly, the remarkable ability of *T. pallidum* to evade clearance from the

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immune system has earned it the designation “stealth pathogen” (106). ~~Also an~~ The activated complement cascade ~~can be seen~~ following spirochete infections (11) ~~which~~ may be used as a non-specific marker of CNS inflammation. Spirochete-host interactions initiate and sustain chronic inflammation ~~triggering various immune responses that activate and end up with various immune responses activating~~ the innate and adaptive immune system, free radicals ~~production~~, apoptosis and amyloid deposition typically seen in AD brains (107).

*P. gingivalis* has been designated as **one of the “keystone” periodontal pathogens** because it is able to establish and maintain the periodontal disease-associated “inflammophilic” microbiota (108). It is able to perform this task as it possesses an awesome variety of virulence factors, recently reviewed by Singhrao et al. (109), to evade the **host immune defenses**, ~~thus~~ serving two major functions: initially ~~for~~ survival of *P. gingivalis* itself via a sustainable inflammatory milieu and ~~then to satisfy its~~ sustainment of nutritional sources by eliminating microbial competitors ~~needs and to stamp out competition~~ (108).

The *P. gingivalis* endotoxin LPS demonstrates differences in the number of phosphate groups together with both the amount of lipid A fatty acids and their specific position. The presence of multiple lipid A structures makes it more difficult for the innate host responses to ~~recognise~~ recognize the molecule thereby aiding the virulence of *P. gingivalis* (110). The consequences of finding *P. gingivalis* LPS in the host’s body, **e.g. the brain (8)**, ~~are~~ include priming of cells for differential activation of the TLR-mediated NF-κB signalling pathway (111) leading to cytokine liberation, complement activation and maintenance of intracerebral inflammation.

*P. gingivalis* evades circulating phagocytes by adhering to erythrocytes (112). An active invasion of *P. gingivalis* and infection-induced complement activation with bystander neural injury was detected in the brains of ApoE<sup>-/-</sup> mice (113). This supported previous notions that bacterial infections can contribute to the development of AD pathology via mechanisms



involving acute phase proteins such as cytokines and the complement cascade where neurons would be attacked.

#### ORAL VIRUS RELATED TO AD

Herpes simplex virus (HSV) is present in more than 70% of the population after 50 years age (114-116). It persists latently in the peripheral nervous system and is periodically reactivated. Characteristically, HSV-1 has been designated as the enemy within (10). Herpes viruses, including Epstein-Barr virus and cytomegalo-virus, are found in high copy counts in aggressive periodontitis, and may interact synergistically with periodontopathic bacteria in the pathogenesis of this disease (117). Periodontal infections activated by Herpes virus ~~Herpes virus active periodontal infections~~ may impair local host defenses and thus increase the aggressiveness of resident periodontopathic bacteria. The bacteria, in turn, may augment the virulence of the herpes viruses.

High proportions of viral-associated proteins in amyloid-containing plaques and/or NFTs corroborate with the involvement of HSV-1 in AD pathology (118). This supports a study by Notably, De Chiara et al. (119) who found reported an association between A $\beta$  accumulation in the brain and HSV infection. Itzhaki et al. (120) suggested that not only does HSV-1 produce the main components of amyloid plaques and NFTs (i.e. A $\beta$  and hyperphosphorylated tau), but it also interferes with the autophagic events that prevent degradation of these proteins and eventually leading to their accumulation in the AD brain. Further, in vitro and in vivo investigations using mouse in murine models following HSV-1 demonstrated A $\beta$  accumulation (121).

A number of scientists have suggested that there is imbalance between production and clearance of  $\beta$ -amyloid in the brain, a ~~thought-premise~~ first proposed by Wisniewski et al.

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on the discovery of soluble species of this protein and later confirmed by Zlokovic et al. (123)  
 (123) ~~to be the case. Thus it~~ It is now widely accepted that defective clearance of this protein  
 brains ~~that leads~~ leading to its accumulation in the form of insoluble A $\beta$ <sub>40/42</sub> plaques.  
 and cytomegalovirus have been detected in the brains of older adults with and without AD  
 (124-126), HSV-1 viral DNA is present in a higher proportion of AD patients (127). It is  
 particularly seen in the temporal and frontal cortices which are the brain regions that are most  
 damaged in AD (128, 129). The relevance of this association is still under investigation;  
 however a plausible role for the HSV-1 viral DNA could be ~~in-associated with~~ the plaque  
 maturation process. Jamieson et al. (127) found that the virus was absent from the brains of  
 most young people, probably because it enters the brain during old age either ~~when the~~  
~~senescence~~ (130) or the virus itself is ~~initially~~ responsible for weakening the host's immune  
 defenses ~~first~~. This latter explanation is likely and is supported by us and others (131).  
 HSV-1 is a strong risk factor for AD in the brains of those with the *APOE $\epsilon$ 4* allele (125, 132).  
 This virus is not only a dormant passenger but can persist in the latent form in neurons or  
 replicate at a very low level in neuroglia (133). During persistence it may release toxic  
 products continuously and induce pro-inflammatory cytokines at low levels which become an  
 additional burden to ~~the a~~ host ~~who is~~ already challenged by age, poor diet, ~~failing-restricted~~  
 exercise as well as any genetic susceptibilities. Itzaki and Wozniak (10) suggested that stress  
 or peripheral infection can reactivate the virus periodically from latency in the brain. This  
 may cause an acute but presumably localized infection, and subsequent damage modulated by  
 the *APOE* gene can lead to formation of A $\beta$  plaques and NFTs.  
 The presence of anti-HSV IgM, a sign of reactivated infection, almost doubled the risk for AD  
 while anti-HSV IgG did not influence the risk (134). Kobayashi et al. (135) suggested that the  
 anti-HSV-1 Ig antibody avidity index could be a useful biomarker for early diagnosis of

anamnesic mild cognitive impairment, which is prodromal to AD, as well as for AD sufferers.

Reactivation of HSV seropositivity is highly correlated with incident-AD (136). Letenneur et al. (136) speculated that AD pathology starts many years before frank dementia and recurrent reactivation of HSV can act as a potent stimulus to brain microglia, increasing cytokine levels, and triggering a positive feedback cycle leading to increasing accumulation of neurohistopathological changes. In other words, infection, followed by local CNS inflammatory reaction is the likely primary ~~occurrence~~ stimulus whereas proteostasis is a consequence of the primary event leading to the development of AD.

Hill et al. (137) suggested a role for HSV-1-induced miRNA-146a in the evasion of HSV-1 from the complement system, ~~which~~ This which is a major first-line host defense mechanism, and the activation of key elements in the arachidonic acid cascade known to contribute to AD-type neuropathological changes.

#### ORAL YEASTS RELATED TO AD

Oral yeast infection ~~is represents~~ a secondary opportunistic infection ~~disease of the diseased~~ ~~where~~ particularly involving *Candida albicans*, but increasingly ~~also~~ *non-albicans* species, e.g. *Candida glabrata* ~~are involved~~. With a growing population of elderly, severe systemic fungal infections have increased dramatically in this age group during the last 30 years (138, 139). Oral yeasts can be found in periodontal pockets, in root canals, on the mucosae and underneath dentures (denture stomatitis) (140-142). Denture stomatitis is prevalent in elderly wearing dentures that are heavily contaminated with yeasts which can be a source of systemic mycosis ~~(Fig. 3)~~. Disseminated mycoses have recently been reported in AD patients (143,

144). Fungal molecules including proteins and polysaccharides [(1,3)- $\beta$ -glucan] were detected in peripheral blood serum, and fungal proteins and DNA were demonstrated by PCR in brain tissue of AD patients. ~~Also chitin~~ Chitin-like fungal structures have also been found in the AD brain (145) and chitinase activity has been proposed as a powerful biomarker of AD (146).

~~Immunohistochemical analyses revealed, albeit in a few cells, in~~ In AD brains ~~containing~~ cytoplasmic material in a small number of cell cells were was targeted by antibodies with immunoreactivity to that immunoreacted with antibodies raised against some yeast cells (147). These findings were consistent with the idea that neurons can be infected by fungi.

Interestingly, antifungal treatment reversed the clinical symptoms of some AD patients (148, 149).

## HOW DO ORAL MICROORGANISMS REACH THE BRAIN?

### Blood stream dissemination

The most likely pathway of for dissemination ~~for of~~ oral microorganisms to the brain is through the blood stream (150). Dental treatment procedures as well as brushing, flossing, chewing and use of tooth picks in a patient with periodontitis will release a bacteraemia (151). This can occur several times during the day and has been estimated to last for up to 3 hours for oral bacteria (152). The bacteraemia is usually ~~taken care of~~ contained by immune cells of the body. However, in people with reduced immune defense, e.g. older individuals, bacteria may ~~settle down within~~ localize to crevices of the oral cavity and vascular channels (150).

### The blood- brain barrier

An intact blood-brain barrier (BBB) prevents microorganisms in the blood from accessing the brain. However, aging favors overgrowth of oral microorganisms, particularly anaerobic bacteria and facultative yeasts that established earlier in life and provoked d pro-inflammatory

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**Commented [PB10]:** also in the general population. Although not on a regular basis, tooth extraction provides a huge bolus of bacteria.

responses that weakened the BBB (16). ~~Actually~~ Notably, magnetic resonance imaging (MRI) confirmed loss of BBB integrity in a mouse model of disseminated candidosis (153). Loss of integrity allows microorganisms to spread through the blood stream and quietly contribute in the pathogenesis of AD. During **immunosenescence**, the innate immune system gradually takes over for the acquired immune system. This contributes to a rise in circulating proinflammatory cytokines such as TNF- $\alpha$  (16). Indeed, proinflammatory mediators can cross the BBB (3, 7, 154). *APOE $\epsilon$ 4*, TNF- $\alpha$  and perhaps Ephrin Type-A Receptor 1 (EphA1) may influence BBB integrity and thus be important for penetration of bacteria, LPS and other toxic bacterial products as well as yeasts into the brains of AD patients (16). *APOE $\epsilon$ 4* affects the integrity of the BBB by activating the cyclophilin A matrix metalloproteinase MM-9 pathway (155).

It is also plausible to suggest that the permeability of the BBB increases with age and thus promotes AD pathogenesis making the brain accessible to microorganisms. Mice with a mutation in the amyloid precursor protein gene which is related to early-onset AD in man, showed increased permeability of the BBB and increased formation of senile plaque as compared to control mice (156). The changes increased with age.

#### Circumventricular organs and perivascular spaces

Circumventricular organs (permit polypeptide hypothalamic hormones to leave the brain without disrupting the BBB) are not dependent on the BBB (56) and may act as another entry portal to the brain for bacteria (157). Poole et al. (8) postulated that bacteria and their products may also directly access the brain via the systemic circulation through the perivascular spaces.

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## The olfactory hypothesis

The “olfactory hypothesis” suggests the olfactory tract as a potential route for pathogenic bacteria to enter the brain and thereby trigger the production of A $\beta$  and NFTs (158). The olfactory and trigeminal nerves are known to be used by periodontal pathogens to bypass the BBB for direct passage to the CNS (5, 150, 159, 160). Identification of oral treponemes in the trigeminal ganglia supports such a route of dissemination (5). Further, ~~s~~Spirochetes may ~~also~~ spread along the fila olfactoria and tractus olfactorius (68, 69).

Olfactory unsheathing cells (OECs) engulf bacteria and migrate towards TNF- $\alpha$  released by activated astrocytes (161). Therefore, OECs could be a vehicle for transporting live bacteria to the brain (i.e., Trojan horse). The olfactory bulb was the first area where NFTs and A $\beta$  deposition were detected in the neuropathological trajectory of AD in humans (162) and in mouse models of AD (163).

## GENETIC, NUTRITIONAL AND ENVIRONMENTAL FACTORS PROMOTING AD

While early-onset AD is genetically determined, LOAD is thought to result from interaction between genetic and environmental factors (12). Several mutated genes are associated with the familial AD, such as the amyloid beta (A $\beta$ ) precursor protein (*A $\beta$ PP*) gene and the presenilin-1 (*PSEN-1*) and *PSEN-2* gene (164-166). A major risk factor for LOAD is polymorphism in the *APO $\epsilon$ 4* allele (2). Also cytokine-related genes seem to be involved in the susceptibility to inflammation in both LOAD (167, 168) and periodontitis (169-171). Thus, polymorphisms that increase TNF- $\alpha$  also increase the risk of both AD and periodontitis (172, 173). Lambert et al. (174) found that 20 different loci can increase host susceptibility to AD including polymorphisms in genes associated with interleukin-1 (IL-1) (71, 175-178) and TNF $\alpha$  (71, 172, 179-181). The *APO $\epsilon$ 4* gene which is one of these 20 loci is highly correlated

with AD (182) but it is also a risk factor for infection and increases the expression of inflammatory mediators (11). Recently, genetic overlap between AD, C-reactive protein (CRP) and plasma lipids was demonstrated by using summary statistics from GWAS of over 200,000 individuals (183). There may also be interplay between genetic risk and environmental risk factors such as toxins and or bacterial, viral and fungal pathogens in LOAD reflecting its complex and multifactorial etiology (1).

Diet with its content of essential B-vitamins, phospholipids and other micronutrients are important for forming new nerve synapses (184). Nutritional deficiencies are common both in elderly and in dementia subjects as briefly discussed by Singhrao et al. (150).

#### ASSOCIATION BETWEEN CHRONIC PERIODONTAL DISEASE AND AD

There is increasing evidence for an association between chronic periodontitis and LOAD (185). Cross-sectional and longitudinal studies have demonstrated that gingival bleeding, loss of periodontal attachment, periodontal probing depth, alveolar bone loss and antibodies to periodontal pathogens are significantly associated with lower cognitive function and decline after adjustment for co-variables (for a review see (12)). Acute phase proteins, including cytokines are possible indirect links between periodontal pathogens and/or their virulence factors (12, 13). Elderly often show neglect of oral hygiene (~~Figs. 3-5~~) which can stimulate recurrent chronic oral infection (150). This again promotes inflammation which can lead to confusion and dementia (3, 4, 154). In 152 subjects 50-70 years of age who were followed for 20 years, greater levels of periodontal inflammation correlated with lower cognitive levels (186). **Furthermore**, gingival bleeding and loss of periodontal attachment ~~apparatus~~ were associated with cognitive impairment in a cohort of 5,138 people aged 20-59 years (187). In 144 nuns, those with encoding APOE $\epsilon$ 4 and who had fewer teeth had experienced more rapid

decline than those with neither or either of these risk factors (188). Clinical and epidemiological studies showed that loss of teeth is associated with poor memory (6, 96, 187, 189). In another study ~~with of~~ 597 community dwelling men followed for 32 years, tooth loss, ~~increasing~~ periodontal pockets ~~depths~~ and progression of alveolar bone loss were associated with impaired cognition particularly in those over 45 years ~~of age~~ (190). Recently, de Souza Rolim et al. (191) found that periodontal infections were more frequent in patients with mild AD than in healthy subjects. Another interesting feature ~~related~~ to the pathogenesis of AD is the low level of infection by “commensals on the loose” (16). These “immuno-tolerated” bacteria may silently multiply in sites outside of their primary niche and an ongoing ~~illness~~ at their secondary location may have significant deleterious effects upon the health of the elderly or demented host with an existing immunocompromised status.

#### PUTATIVE TREATMENT AND PROPHYLAXIS OF AD

There is no effective treatment or prophylaxis yet for AD, but several approaches have been proposed. Efforts in this respect are important. If we could delay onset of dementia by only 2 years we might lower the prevalence of AD by more than 22 million cases over the next 40 years (14). ~~Indeed, delaying the disease process is a better option as the~~ ~~Notabley~~Notably, the of the *APOEε4* allele in the very old (90+) age group, appears to confer protection (192), having bypassed a period of being at risk around 85+ years of age.

If periodontal disease is implicated in AD, periodontitis prophylaxis ~~should be feasible~~ ~~could~~ ~~be of help~~. It would be interesting to see if this has any effect on the initiation and aggravation of AD but an observation period of decennia is probably needed.

~~In a study of subjects with mild to moderate AD, a~~ 3-month course of doxycycline and rifampicin reduced cognitive deterioration ~~in during~~ a 6 months' ~~follow-up~~ ~~follow-up~~ interval

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study in subjects with mild to moderate AD (193). It was concluded that use of antibacterial the treatment of *C. pneumoniae* but had a beneficial effect on cognitive decline in AD (193). This might be related to prevention or attenuation of a number of peripheral infections or dampening down the proinflammatory cytokine response. ~~Minicycline~~Minocycline was found early, pre-plaque neuroinflammation and inhibit the APP cleaving enzyme 1 (BACE-1) in a transgenic model of Alzheimer's disease-like amyloid pathology (194). It was suggested that interfering with inflammation could be a useful therapeutic approach in early, pre-plaque stages of AD-like amyloid pathology.

Anti-inflammatory drugs given for at least 2 years before the onset of dementia delayed the disease process (~~194195-196~~197). It may also be beneficial to combine anti-inflammatory antibacterials (193). Examination of several available Non-steroidal Anti-Inflammatory Drugs (NSAIDs) showed that only a few of them had any useful A $\beta$ -modifying or other activity of therapeutic use in LOAD (for a review see (1)).

Itzhaki and Wozniak (10, ~~197~~198) suggested that antiviral therapy and perhaps vaccination against HSV-1 in early life could be useful. If HSV-1 is implicated in AD, vaccination could prevent the excessive accumulation of A $\beta$  in the brain. Vaccination with mixed HSV glycoproteins prior to HSV infection protected against viral latency in mouse brains (~~198~~199). Also Mori (~~199~~200) maintained that antiviral approaches including chemotherapy and vaccination are promising for prevention and treatment of AD and remain to be validated. Furthermore, Carter (118) suggested that vaccination or antiviral agents and immune suppressants may be considered as therapeutic options before or ~~in~~-during the early stages of AD. Interestingly, exposure of HSV-1-infected cell cultures to intravenous immunoglobulin acting via anti- $\beta$ -amyloid ~~antibodies~~antibodies reduced the accumulation of A $\beta$  and phosphorylated tau (~~200~~201).

Angiotensin-converting enzyme (ACE) from *Stigmatella aurantiaca* may cleave the A $\beta$  peptide similar to human ACE and may be used as a novel form of treatment against AD (201202). Furthermore, Chiarini et al. (202203) maintained that calcilytics could halt AD progression and preserve the patients' cortical neurons, cognitive abilities, and eventually life if given at minimal cognitive impairment or at earlier stages. Studies ~~from~~ using mice suggested the use of tau aggregation inhibitors as potential drugs for the treatment of AD and other tauopathies (203204).

Resveratrol is a polyphenol present in red wine. Its capability of directly interfering with the toxic  $\beta$ -amyloid protein aggregation in AD has recently been shown (204205). Resveratrol was found to reduce A $\beta$ -induced toxicity in a *Caenorhabditis elegans* model of AD by targeting specific proteins involved in proteostasis and thereby reducing the amount of aggregated A $\beta$  (205206). This is in concert with our previous finding that the effect of a drinking pattern of 2-7 times per week reduced the risk of myocardial infarction among men who had a history of tooth extractions due to periodontal/dental infection (206207).

Potent inhibitors of A $\beta$  oligomer formation or A $\beta$ -induced cell toxicity have proven to be attractive means for therapeutic intervention of AD. Song et al. (207208) found that the anti-Alzheimer effects of centipedegrass, which contains several C-glycosyl flavone constituents, occurred through inhibition of neuronal cell death by intervening with oligomeric A $\beta$  formation and reducing beta-site amyloid precursor protein cleaving enzyme 1 (BACE1) activity. The authors suggested that Maysin, a major flavonoid of corn silk, in centipedegrass could be an excellent therapeutic candidate for the prevention of AD.

Active immunization against important domains of Alzheimer tau eliminated tau aggregation and neurofibrillary pathology (208209). The AD type of tau hyperphosphorylation was abolished in transgenic mice by vaccination across a wide range of AD phospho-epitopes.

Kontseikova et al. (208209) demonstrated that active immunization of rats with a tau peptide encompassing the epitope revealed by monoclonal antibody DC8E8 led to elimination of all major hallmarks of neurofibrillary pathology involving a 95% reduction in the AD-type hyperphosphorylation of tau.

## CONCLUSIONS

LOAD which is the predominant form of AD, does not seem to have a single cause. On the contrary, a multitude of factors may be involved and they may act in concert. ~~Of these~~ Among others, both genetic and environmental factors may be involved. Even among cooperation, action may occur since the brain can hardly differentiate between different microbial insults which collectively contribute capacity for enhancing all end up in Irrespective of the cause, systemic inflammation may predict the onset of dementia. Organisms such as spirochetes, *P. gingivalis*, *C. pneumoniae*, *H. pylori*, ~~H~~ herpes simplex type virus and *Candida* are among the prime candidate pathogens the most suspected pathogens in events causing AD, oral microorganisms may play a role, particularly anaerobic bacteria such as treponemes, *P. gingivalis*, *Prevotella* spp., *Fusobacterium* and *Actinomyces*, but also facultative anaerobic *Candida* species. It is important to recognize that infection can occur decades before the manifestation of dementia. The most convincing evidence for a causal relationship between oral bacteria and AD is that noted for spirochetes which are both neurotropic and motile. ~~They also fulfill Koch's and Hill's postulates for a causal relationship.~~ It is likely that oral infection can be a risk factor for Alzheimer's disease but it is not the only one. Experiments in humans in vivo may require long exposure times to disclose key events and mechanisms of AD. There is, as yet, no cure for AD despite concerted efforts and investment by industry and this is not without concerted efforts from investment by industry but because drug discovery in dementia is hugely challenging. Prevention of AD through

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long-term use of antibiotics may be impractical and could select for resistant bacteria. This is worrisome as the prevalence of AD and the public expenses related to its management are expected to increase greatly in the next **decade**.

in AD, ~~then~~ dental hygiene and treatment will provide the AD prophylaxis from an early age ~~this oral disease-periodontitis~~ is modifiable. However, improving oral hygiene and treating in the AD patient can be challenging since patients are often uncooperative.- There is also for training care-givers to assist with oral care in such patients.

Vaccination against key organisms and important domains of AD has had some beneficial effect. Also several agents interfering directly with the pathogenesis of AD have been tested. In order to find a cure, there is a need for clinical diagnostic information and knowledge of the causal agents for AD ~~AD-causative agents~~ so that specific treatment options targeting these organisms ~~against these organisms~~ can be developed. As for diagnostic biomarkers, increased antibody levels to specific oral pathogens in particular to *P. gingivalis* may be used as a preventive-monitoring tool years before clinical manifestation of AD. This is important because treatment will probably have to start early.

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## CONFLICT OF INTEREST AND FUNDING

There is no conflict of interest in the present study for any of the authors. Funding was as given under Acknowledgement.

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## REFERENCES

1. Balin BJ, Hudson AP. Etiology and pathogenesis of late-onset Alzheimer's disease. *Curr Allergy Asthma Rep* 2014; 14: 417. doi 10.1007/s11882-013-0417-1.
2. Corder EH, Saunders AM, Strittmatter WJ, Schmechel DE, Gaskell PC, Small GW, et al. Gene dose of apolipoprotein E type 4 allele and the risk of Alzheimer's disease in late onset families. *Science* 1993; 261: 921-3.
3. Holmes C, El-Okl M, Williams AL, Cunningham C, Wilcockson D, Perry VH. Systemic infection, interleukin 1beta, and cognitive decline in Alzheimer's disease. *J Neurol Neurosurg Psychiatry* 2003; 74: 788-9.
4. Dunn N, Mullee M, Perry VH, Holmes C. Association between dementia and infectious disease: evidence from a case-control study. *Alzheimer Dis Assoc* 2005; 19: 91-4.
5. Riviere GR, Riviere KH, Smith KS. Molecular and immunological evidence of oral *Treponema* in the human brain and their association with Alzheimer's disease. *Oral Microbiol Immunol* 2002; 17: 113-8.
6. Stein PS, Desrosiers M, Donegan SJ, Yepes JF, Kryscio RJ. Tooth loss, dementia and neuropathy in the NUN study. *J Am Dent Assoc* 2007; 138: 1314-22; quiz 1381-2.
7. Kamer AR, Craig RG, Pirraglia E, Dasanayake AP, Norman RG, Boylan RJ, et al. TNF- $\alpha$  and antibodies to periodontal bacteria discriminate between Alzheimer's disease patients and normal subjects. *J Neuroimmunol* 2009; 216: 92-7.
8. Poole S, Singhrao SK, Kesavalu L, Curtis MA, Crean S. Determining the presence of periodontopathic virulence factors in short-term postmortem Alzheimer's disease brain tissue. *J Alzheimers Dis* 2013; 36: 665-77. doi: 10.3233/JAD-121918.

9. Balin BJ, Little CS, Hammond CJ, Appelt DM, Whittum-Hudson JA, Gérard HC, et al.  
*Chlamydomydia pneumoniae* and the etiology of late-onset Alzheimer's disease. J  
 Alzheimer's Dis 2008; 371-80.
10. Itzhaki RF, Wozniak MA. Herpes simplex virus type 1 in Alzheimer's disease: the  
 enemy within. J Alzheimer's Dis 2008; 13: 393-405.
11. Miklossy J. Chronic inflammation and amyloidogenesis in Alzheimer's disease – role of  
 spirochetes. J Alzheimer's Dis 2008; 13: 381-91.
12. Kamer AR, Dasanayake AP, Craig RG, Glodzik-Sobanska L, Bry M, de Leon MJ.  
 Alzheimer's disease and peripheral infections: the possible contribution from  
 periodontal infections, model and hypothesis. J Alzheimer's Dis 2008; 13: 437-49.
13. Watts A, Crimmins EM, Gatz M. Inflammation as a potential mediator for the  
 association between periodontal disease and Alzheimer's disease. Neuropsychiatr Dis  
 Treat 2008; 4: 865-76.
14. Brookmeyer R, Johnson E, Ziegler-Graham K, Arrighi HM. Forecasting the global  
 burden of Alzheimer's disease. Alzheimers Dement 2007; 3: 186-91.
15. Ouerfurth HW, LaFerla FM. Alzheimer's disease. New Engl J Med 2010; 362: 329-44.
16. Shoemark DK, Allen SJ. The microbiome and disease: reviewing the links between the  
 oral microbiome, aging and Alzheimer's disease. J Alzheimer's Disease 2015; 43: 725-  
 38.
17. Alzheimer A. Über eine eigenartige Erkrankung der Hirnrinde. All Z Psychiat 1907; 64:  
 146-8.
18. Selkoe DJ. Alzheimer's disease. Cold Spring Harb Perspect Biol 2011; 2011; 3. pii:  
 a004457. doi: 10.1101/cshperspect.a004457.

19. Yankner BA, Dawes LR, Fisher S, Villa-Komaroff L, Oster-Granite ML, Neve RL. Neurotoxicity of a fragment of the amyloid precursor associated with Alzheimer's disease. *Science* 1989; 245: 417-20.
20. Deshpande A, Mina E, Glabe C, Busciglio J. Different conformations of amyloid beta induce neurotoxicity by distinct mechanisms in human cortical neurons. *J Neurosci* 2006; 26: 6011-8.
21. Glabe CC. Amyloid accumulation and pathogenesis of Alzheimer's disease: significance of monomeric, oligomeric and fibrillar A $\beta$ . *Subcell Biochem* 2005; 38: 167-77.
22. Shankar GM, Li S, Mehta TH, Garcia-Munoz A, Shepardson NE, Smith I, et al. Amyloid-beta protein dimers isolated directly from Alzheimer's brains impair synaptic plasticity and memory. *Nat Med* 2008; 14: 837-42. doi: 10.1038/nm1782.
23. Soscia SJ, Kirby JE, Washicosky KJ, Tucker SM, Ingelsson M, Hyman B, et al. The Alzheimer's disease-associated amyloid beta-protein is an antimicrobial peptide. *PloS One* 2010; 5, e9505. doi: 10.1371/journal.pone.0009505.
24. Akiyama H, Barger S, Barnum S, Bradt B, Bauer J, Cole GM, et al. Inflammation and Alzheimer's disease. *Neurobiol Aging* 2000; 21: 383-421.
25. Perry VH, Nicoll JA, Holmes C. Microglia in neurodegenerative disease. *Nat Rev Neurol* 2010; 6: 193-201. doi: 10.1038/nrneurol.2010.17.
26. Hanisch UK. Microglia as a source and target of cytokines. *Glia* 2002; 40: 140-55.
27. Lacroix S, Feinstein D, Rivest S. The bacterial endotoxin lipopolysaccharide has the ability to target the brain in upregulating its membrane CD14 receptor within specific cellular populations. *Brain Pathol* 1998; 8: 625-40.
28. Laflamme N, Rivest S. Toll-like receptor 4: the missing link of the cerebral innate immune response triggered by circulating gram-negative bacterial cell wall components. *FASEB J* 2001; 15: 155-63.



29. Beutler B, Hoebe K, Du X, Ulevitch RJ. How we detect microbes and respond to them: the Toll-like receptors and their transducers. *J Leukoc Biol* 2003; 74: 479-85.
30. Rivest S. Regulation of innate immune responses in the brain. *Nat Rev Immunol* 2009; 9: 429-39. doi: 10.1038/nri2565.
31. Iqbal K, Grue-Iqbal I. Ubiquitination and abnormal phosphorylation of paired helical filaments in Alzheimer's disease. *Mol Neurobiol* 1991; 5: 399-410.
32. Malpass K. Alzheimer disease: functional dissection of CD33 locus implicates innate immune response in Alzheimer disease pathology. *Nat Rev Neurol* 2013; 9: 360. doi: 10.1038/nrneurol.2013.119.
33. Shulman JM, Chen K, Keenan BT, Chibnik LB, Fleisher A, Thiyyagura P, et al. Genetic susceptibility for Alzheimer disease neuritic plaque pathology. *JAMA Neurol* 2013 1;70: 1150-7. doi: 10.1001/jamaneurol.2013.2815.
34. Guerreiro RJ, Hardy J. Alzheimer's disease genetics: lessons to improve disease modelling. *Biochem Soc Trans* 2011; 39:910-6. doi: 10.1042/BST0390910.
35. Terry RD. Physical basis of cognitive alterations in Alzheimer's disease: synapse loss is the major correlate of cognitive impairment. *Ann Neurol* 1991; 30: 572-80.
36. Masliah E, Mallory M, Hansen L, Alford M, Albright T, DeTeresa R, et al. Patterns of aberrant sprouting in Alzheimer's disease. *Neuron* 1991; 6: 729-39.
37. Masliah E, Mallory M, Hansen L, DeTeresa R, Terry RD. Quantitative synaptic alterations in the human neocortex during normal aging. *Neurology* 1993; 43: 192-7.
38. Kondo K, Niino M, Shido K. A case-control study of Alzheimer's disease in Japan--significance of life-styles. *Dementia* 1994; 5: 314-26.
39. Bartus RT, Dean RL 3rd, Beer B, Lippa AS. The cholinergic hypothesis of geriatric memory dysfunction. *Science* 1982; 217: 408-14.

- 741 40. Demetrius LA, Magistretti PJ, Pellerin L. Alzheimer's disease: the amyloid hypothesis  
742 and the Inverse Warburg effect. *Front Physiol* 2015; 5:522. doi:  
743 10.3389/fphys.2014.00522.
- 744 41. Tanzi RE, Watkins PC, Stewart GD, Wexler NS, Gusella JF, Haines JL. A genetic  
745 linkage map of human chromosome 21: analysis of recombination as a function of sex  
746 and age. *Am J Hum Genet* 1992; 50: 551-8.
- 747 42. Scheuner D, Eckman C, Jensen M, Song X, Citron M, Suzuki N, et al. Secreted amyloid  
748 beta-protein similar to that in the senile plaques of Alzheimer's disease is increased *in*  
749 *vivo* by the presenilin 1 and 2 and APP mutations linked to familial Alzheimer's disease.  
750 *Nat Med* 1996; 2:864-70.
- 751 43. Glenner GG, Wong CW. Alzheimer's disease and Down's syndrome: sharing of a unique  
752 cerebrovascular amyloid fibril protein. *Biochem Biophys Res Commun* 1984; 122:  
753 1131-5.
- 754 44. Lambert JC, Heath S, Even G, Campion D, Sleegers K, Hiltunen M, et al. Genome-wide  
755 association study identifies variants at CLU and CR1 associated with Alzheimer's  
756 disease. *Nat Genet* 2009; 41: 1094-9. doi: 10.1038/ng.439.
- 757 45. Thambisetty M, An Y, Nalls M, Sojkova J, Swaminathan S, Zhou Y, et al. Effect of  
758 complement CR1 on brain amyloid burden during aging and its modification by APOE  
759 genotype. *Biol Psychiatry* 2013; 73: 422-8. doi: 10.1016/j.biopsych.2012.08.015.
- 760 46. Killick R, Hughes TR, Morgan BP, Lovestone S. Deletion of Crry, the murine ortholog  
761 of the sporadic Alzheimer's disease risk gene CR1, impacts tau phosphorylation and  
762 brain CFH. *Neurosci Lett* 2013; 533: 96-9. doi: 10.1016/j.neulet.2012.11.008.
- 763 47. Jack CR Jr, Knopman DS, Jagust WJ, Shaw LM, Aisen PS, Weiner MW, et al.  
764 Hypothetical model of dynamic biomarkers of the Alzheimer's pathological cascade.  
765 *Lancet Neurol* 2010; 9: 119-28. doi: 10.1016/S1474-4422(09)70299-6.

48. Morgan BP, Gasque P. Expression of complement in the brain: role in health and disease. *Immunol Today* 1996; 17: 461-6.
49. Benveniste EN. Cytokine actions in the central nervous system. *Cytokine Growth Factor Rev* 1998; 9: 259-75.
50. Gasque P. Complement: a unique innate immune sensor for danger signals. *Mol Immunol* 2004; 41: 1089-98.
51. Harold D, Abraham R, Hollingworth P, Sims R, Gerrish A, Hamshere ML, et al. Genome-wide association study identifies variants at CLU and PICALM associated with Alzheimer's disease. *Nat Genet* 2009; 41: 1088-93. doi: 10.1038/ng.440. Erratum in: *Nat Genet* 2009; 41: 1156. *Nat Genet* 2013; 45: 712. Haun, Reinhard [added].
52. Eikelenboom P, Stam FC. Immunoglobulins and complement factors in senile plaques. An immunoperoxidase study. *Acta Neuropathol* 1982; 57: 239-42.
53. McGeer PL, Akiyama H, Itagaki S, McGeer EG. Activation of the classical complement pathway in brain tissue of Alzheimer patients. *Neurosci Lett* 1989; 107: 341-6.
54. Rogers J, Cooper NR, Webster S, Schultz J, McGeer PL, Styren SD, et al. Complement activation by beta-amyloid in Alzheimer disease. *Proc Natl Acad Sci U S A* 1992; 89: 10016-20.
55. Shen Y, Lue L, Yang L, Roher A, Kuo Y, Strohmeier R, et al. Complement activation by neurofibrillary tangles in Alzheimer's disease. *Neurosci Lett* 2001; 305: 165-8.
56. Oldfield BJ, Mckinley MJ. Circumventricular organs. In *The Rat Nervous System*. Paxinos G, ed. Academic Press, San Diego 1995, pp 391-403.
57. Holmes C, Cunningham C, Zotova E, Woolford J, Dean C, Kerr S, et al. Systemic inflammation and disease progression in Alzheimer disease. *Neurology* 2009; 73: 768-74. doi: 10.1212/WNL.0b013e3181b6bb95.

58. de Oliveira JM, Lisboa Lde B. Hospital-acquired infections due to gram-negative bacteria. *N Engl J Med* 2010; 363: 1482-3; author reply 1483-4.
59. Honjo K, van Reekum R, Verhoeff NPLG. Alzheimer's disease and infection: Do infectious agents contribute to progression of Alzheimer's disease? *Alzheimers Dement* 2009; 5: 348-60. doi: 10.1016/j.jalz.2008.12.001.
60. Maheshwari P, Eslick GD. Bacterial infection and Alzheimer's disease: a meta-analysis. *J Alzheimer's Dis* 2015; 43: 957-66.
61. Shima K, Kuhlenbäumer G, Rupp J. *Chlamydia pneumoniae*-infection and Alzheimer's disease: a connection to remember? *Med Microbiol Immunol* 2010; 199: 283-9. doi: 10.1007/s00430-010-0162-1.
62. Lim C, Hammond CJ, Hingley ST, Balin BJ. *Chlamydia pneumoniae* infection of monocytes *in vitro* stimulates innate and adaptive immune responses relevant to those in Alzheimer's disease. *J Neuroinflammation* 2014; 11: 217. doi: 10.1186/s12974-014-0217-0.
63. Hammond CJ, Hallock LR, Howanski RJ, Appelt DM, Little CS. Immunohistological detection of *Chlamydia pneumoniae* in the Alzheimer's disease brain. *Neuroscience* 2010; 11: 121.
64. Hogan RJ, Mathews SA, Mukhopadhyay S, Summersgill JT, Timms P. Chlamydial persistence: beyond the biphasic paradigm. *Infect Immun* 2004; 72: 1843-55.
65. Whittum-Hudson JA, Schumacher HR, Hudson AP. *Chlamydia pneumoniae* and inflammatory arthritis. In: Yamamoto Y, Friedman H, Bendinelli M, editors. *Chlamydia pneumoniae* infection and diseases. New York: Kluwer/Academic Press; 2004. p. 227-38.
66. Dreses-Werringloer U, Bhuiyan M, Zhao Y, Gérard HC, Whittum-Hudson JA, Hudson AP. Initial characterization of *Chlamydophila (Chlamydia) pneumoniae* cultured from the late-onset Alzheimer brain. *Int J Med Microbiol* 2009; 299: 187-201.

67. Dezfulian M, Shokrgozar MA, Sardari S, Parivar K, Javadi G. Can phages cause Alzheimer's disease? *Med Hypotheses* 2008; 71: 651-6. doi: 10.1016/j.mehy.2008.07.005.
68. Miklossy J. Alzheimer's disease - a neurospirochetosis. Analysis of the evidence following Koch's and Hill's criteria. *J Neuroinflammation* 2011; 8: 90. doi: 10.1186/1742-2094-8-90.
69. Miklossy J. Emerging roles of pathogens in Alzheimer disease. *Expert Rev Mol Med* 2011; 13:e30. doi: 10.1017/S1462399411002006.
70. Ohnishi S, Koide A, Koide S. Solution conformation and amyloid-like fibril formation of a polar peptide derived from a beta-hairpin in the OspA single-layer beta-sheet. *J Mol Biol* 2000; 301: 477-89.
71. McGeer PL, McGeer EG. Polymorphisms in inflammatory genes and the risk of Alzheimer disease. *Arch Neurol* 2001; 58: 1790-2.
72. Miklossy J. Historic evidence to support a causal relationship between spirochetal infections and Alzheimer's disease. *Front Aging Neurosci* 2015; 7:46. doi: 10.3389/fnagi.2015.00046.
73. Fallon BA, Nields JA. Lyme disease: a neuropsychiatric illness. *Am J Psychiatry* 1994 ; 151: 1571-83.
74. MacDonald AB, Miranda JM. Concurrent neocortical borreliosis and Alzheimer's disease. *Hum Pathol* 1987; 18: 759-61.
75. MacDonald AB. Concurrent neocortical borreliosis and Alzheimer's Disease. Demonstration of a spirochetal cyst form. *Ann N Y Acad Sci* 1988; 539, 468-70. doi: 10.1111/j.1749-6632.1988.tb31909.x
76. MacDonald AB. Transfection "Junk" DNA - a link to the pathogenesis of Alzheimer's disease? *Med Hypotheses* 2006; 66:1140-1.

77. Miklossy J, Khalili K, Gern L, Ericson RL, Darekar P, Bolle L, et al. *Borrelia burgdorferi* persists in the brain in chronic lyme neuroborreliosis and may be associated with Alzheimer disease. *J Alzheimers Dis* 2004; 6: 639-49; discussion 673-81.
78. Bu XL, Yao XQ, Jiao SS, Zeng F, Liu YH, Xiang Y, et al. A study on the association between infectious burden and Alzheimer's disease. *Eur J Neurol* 2014. doi: 10.1111/ene.12477.
79. Gutacker M, Valsangiacomo C, Balmelli T, Bernasconi MV, Bouras C, Piffaretti JC. Arguments against the involvement of *Borrelia burgdorferi* sensu lato in Alzheimer's disease. *Res Microbiol* 1998; 149: 31-7.
80. Pappolla MA, Omar R, Saran B, Andorn A, Suarez M, Pavia C, et al. Concurrent neuroborreliosis and Alzheimer's disease: analysis of the evidence. *Hum Pathol* 1989; 20: 753-7.
81. Shiota S, Murakami K, Yoshiiwa A, Yamamoto K, Ohno S, Kuroda A, et al. The relationship between *Helicobacter pylori* infection and Alzheimer's disease in Japan. *J Neurol* 2011; 258: 1460-3. doi: 10.1007/s00415-011-5957-5.
82. Kountouras J, Zavos C, Boziki M, Gavalas E, Kyriakou P, Deretzi G, et al. Association between *Helicobacter pylori* infection and Alzheimer's disease in Japan. *J Neurol* 2011; 258: 2086. doi: 10.1007/s00415-011-6054-5.
83. Kountouras J, Boziki M, Gavalas E, Zavos C, Deretzi G, Chatzigeorgiou S, et al. Five-year survival after *Helicobacter pylori* eradication in Alzheimer disease patients. *Cogn Behav Neurol* 2010; 23: 199-204. doi: 10.1097/WNN.0b013e3181df3034.
84. Aas JA, Paster BJ, Stokes LN, Olsen I, Dewhirst FE. Defining the normal bacterial flora of the oral cavity. *J Clin Microbiol* 2005; 43: 5721-32.
85. Dewhirst FE, Chen T, Izard J, Paster BJ, Tanner AC, Yu WH, et al. The human microbiome. *J Bacteriol* 2010; 192: 5002-17. doi: 10.1128/JB.00542-10.

86. Imangaliyev S, Keijser B, Crielaard W, Tsivtsivadze E. Personalized microbial network inference via co-regularized spectral clustering. *Methods* 2015. pii: S1046-2023(15)00123-1. doi: 10.1016/j.ymeth.2015.03.017.
87. Segata N, Haake SK, Mannon P, Lemon KP, Waldron L, Gevers D, et al. Composition of the adult digestive tract bacterial microbiome based on seven mouth surfaces, tonsils, throat and stool samples. *Genome Biol* 2012; 13: R42. doi: 10.1186/gb-2012-13-6-r42.
88. Axelsson P, Lindhe J, Nyström B. On the prevention of caries and periodontal disease. Results of a 15-year longitudinal study in adults. *J Clin Periodontol* 1991; 18: 182-9.
89. Flemmig TF. Periodontitis. *Ann Periodontol* 1999; 4: 32-8.
90. Armitage GC. Development of a classification system for periodontal diseases and conditions. *Ann Periodontol* 1999; 4: 1-6.
91. Holt SC, Ebersole JL. *Porphyromonas gingivalis*, *Treponema denticola*, and *Tannerella forsythia*: the "red complex", a prototype polybacterial pathogenic consortium in periodontitis. *Periodontol 2000* 2005; 38: 72-122.
92. Colombo AP, Boches SK, Cotton SL, Goodson JM, Kent R, Haffajee AD, et al. Comparisons of subgingival microbial profiles of refractory periodontitis, severe periodontitis, and periodontal health using the human oral microbe identification microarray. *J Periodontol* 2009; 80: 1421-32. doi: 10.1902/jop.2009.090185.
93. Preza D, Olsen I, Willumsen T, Boches SK, Cotton SL, Grinde B, et al. Microarray analysis of the microflora of root caries in elderly. *Eur J Clin Microbiol Infect Dis* 2009; 28: 509-17. doi: 10.1007/s10096-008-0662-8.
94. Torlakovic L, Klepac-Ceraj V, Ogaard B, Cotton SL, Paster BJ, Olsen I. Microbial community succession on developing lesions on human enamel. *J Oral Microbiol* 2012; 4. doi: 10.3402/jom.v4i0.16125.
95. Hardy JA, Mann DM, Wester P, Winblad B. An integrative hypothesis concerning the

- pathogenesis and progression of Alzheimer's disease. *Neurobiol Aging* 1986; 7: 489-502.
96. Noble JM, Borrell LN, Papapanou PN, Elkind MSV, Scarmeas N, Wright CB. Periodontitis is associated with cognitive impairment among older adults: Analysis of NHANES-III. *J Neurol Neurosurg Psychiatry* 2009; 80: 1206-11. doi:10.1136/jnp.2009.174029.
97. Sparks Stein P, Steffen MJ, Smith C, Jicha G, Ebersole JL, Abner E, et al. Serum antibodies to periodontal pathogens are a risk factor for Alzheimer's disease. *Alzheimers Dement* 2012; 8: 196-203. doi: 10.1016/j.jalz.2011.04.006.
98. Noble JM, Scarmeas N, Celenti RS, Elkind MSV, Wright CB, Schupf N, et al. Serum IgG antibody levels to periodontal microbiota are associated with incident Alzheimer disease. *PLoS One* 2014; 9: e114959. doi: 10.1371/journal.pone.0114959.
99. Hajishengallis G. Too old to fight? Aging and its toll on innate immunity. *Mol Oral Microbiol* 2010; 25: 25-37. doi: 10.1111/j.2041-1014.2009.00562.x.
100. Ricklin D, Hajishengallis G, Yang K, Lambris JD. Complement: a key system for immune surveillance and homeostasis. *Nat Immunol* 2010; 11: 785-97. doi: 10.1038/ni.1923.
101. Fong ON, Chan KY, Leung KT, Lam HS, Cheung HM, Leung TY, et al. Expression profile of cord blood neutrophils and dysregulation of HSPA1A and OLR1 upon challenge by bacterial peptidoglycan. *J Leukoc Biol* 2014; 95: 169-78. doi: 10.1189/jlb.0413219.
102. Bibi F, Yasir M, Sohrab SS, Azhar EI, Al-Qahtani MH, Abuzenadah AM, et al. Link between chronic bacterial inflammation and Alzheimer disease. *CNS Neurol Disord Drug Targets* 2014; 13: 1140-7.
103. Miklossy J, Kraftsik R, Pillevuit O, Lepori D, Genton C, Bosman FT. Curly fiber and



tangle-like inclusions in the ependyma and choroid plexus--a pathogenetic relationship with the cortical Alzheimer-type changes? J Neuropathol Exp Neurol 1998; 7: 1202-12.

104. Miklossy J, Kis A, Radenovic A, Miller L, Forro L, Martins R, et al. Beta-amyloid deposition and Alzheimer's type changes induced by *Borrelia* spirochetes. Neurobiol Aging 2006; 27: 228-36.

105. Hachinsky V, Munoz DG. Cerebrovascular pathology in Alzheimer's disease: cause, effect or epiphenomenon? Ann N Y Acad Sci 1997; 826:1-6.

106. Radolf JD, Desrosiers DC. *Treponema pallidum*, the stealth pathogen, doth change, but how? Mol Microbiol 2009; 72: 1081-6. doi: 10.1111/j.1365-2958.2009.06711.x.

107. Snowdon DA, Greiner LH, Mortimer JA, Riley KP, Greiner PA, Markesbery WR. Brain infarction and the clinical expression of Alzheimer disease. The Nun Study. JAMA 1997; 277: 813-7.

108. Hajishengallis G. The inflammophilic character of the periodontitis-associated microbiota. Mol Oral Microbiol 2014; 29: 248-57. doi: 10.1111/omi.12065.

109. Singhrao SK, Harding A, Poole S, Kesavalu L, Crean S. *Porphyromonas gingivalis* periodontal infection and its putative links with Alzheimer's disease. Mediators Inflamm 2015; 2015:137357.

110. Reife RA, Coats SR, Al-Qutub M, Dixon DM, Braham PA, Billharz RJ, et al. *Porphyromonas gingivalis* lipopolysaccharide lipid A heterogeneity: differential activities of tetra- and penta-acylated lipid A structures on E-selectin expression and TLR4 recognition. Cell Microbiol 2006; 8: 857-68.

111. Kocgozlu L, Elkaim R, Tenenbaum H, Werner S. Variable cell responses to *P. gingivalis* lipopolysaccharide. J Dent Res 2009; 88: 741-5. doi: 10.1177/0022034509341166.

112. Belstrøm D, Holmstrup P, Damgaard C, Borch TS, Skjødt MO, Bendtzen K, et al. The atherogenic bacterium *Porphyromonas gingivalis* evades circulating phagocytes by adhering to erythrocytes. *Infect Immun* 2011; 79: 1559-65. doi: 10.1128/IAI.01036-10.
113. Poole S, Singhrao SK, Chukkapalli S, Rivera M, Velsko I, Kesavalu L, et al. Active invasion of *Porphyromonas gingivalis* and infection-induced complement activation in ApoE<sup>-/-</sup> mice brains. *J Alzheimer's Dis* 2015; 43: 67-80.
114. Xu F, Sternberg MR, Kottiri BJ, McQuillan GM, Lee FK, Nahmias AJ, et al. Trends in herpes simplex virus type 1 and type 2 seroprevalence in the United States. *JAMA* 2006; 296: 964-73.
115. Bünzli D, Wietlisbach V, Barazzoni F, Sahli R, Meylan PR. Seroepidemiology of Herpes simplex virus type 1 and 2 in Western and Southern Switzerland in adults aged 25-74 in 1992-93: a population-based study. *BMC Infect Dis* 2004; 17: 10.
116. Malkin JE, Morand P, Malvy D, Ly TD, Chanzy B, de Labareyre C, et al. Seroprevalence of HSV-1 and HSV-2 infection in the general French population. *Sex Transm Infect* 2002; 78:201-3.
117. Slots J. Herpesvirus periodontitis: infection beyond biofilm. *J Calif Dent Assoc* 2011; 39: 393-9.
118. Carter CJ. Alzheimer's disease plaques and tangles: Cemeteries of a Pyrrhic victory of the immune defense network against herpes simplex infection at the expense of complement and inflammation-mediated neuronal destruction. *Neurochem Int* 2011; 58: 301-20. doi: 10.1016/j.neuint.2010.12.003.
119. De Chiara G, Marcocci ME, Civitelli L, Argnani R, Piacentini R, Ripoli C, et al. APP processing induced by herpes simplex virus type 1 (HSV-1) yields several APP fragments in human and rat neuronal cells. *PLoS One* 2010; 5: e13989. doi:

- 10.1371/journal.pone.0013989.
120. Itzhaki RF, Cosby SL, Wozniak MA. Herpes simplex virus type 1 and Alzheimer's disease: the autophagy connection. *J Neurovirol* 2008; 14: 1-4. doi: 10.1080/13550280701802543.
121. Wozniak MA, Mee AP, Itzhaki RF. Herpes simplex virus type I DNA is located within Alzheimer's disease amyloid plaques. *J Pathol* 2009; 217: 131-8.
122. Wisniewski T, Ghiso J, Frangione B. Alzheimer's disease and soluble A beta. *Neurobiol Aging* 1994; 15:143-52.
123. Zlokovic BV, Yamada S, Holtzman D, Ghiso J, Frangione B. Clearance of amyloid beta-peptide from brain: transport or metabolism? *Nat Med* 2000; 6:718-9.
124. Aiello AE, Haan M, Blythe L, Moore K, Gonzalez JM, Jagust W. The influence of latent viral infection on rate of cognitive decline over 4 years. *J Am Geriatr Soc* 2006; 54: 1046-54.
125. Itzhaki RF, Lin WR, Shang D, Wilcock GK, Faragher B, Jamieson GA. Herpes simplex virus type 1 in brain and risk of Alzheimer's disease. *Lancet* 1997; 349: 241-4.
126. Lurain NS, Hanson BA, Martinson J, Leurgans SE, Landay AL, Bennett DA, Schneider JA. Virological and immunological characteristics of human cytomegalovirus infection associated with Alzheimer disease. *J Infect Dis* 2013; 208: 564-72. doi: 10.1093/infdis/jit210.
127. Jamieson GA, Maitland NJ, Wilcock GK, Yates CM, Itzhaki RF. Herpes simplex virus type 1 DNA is present in specific regions of brain from aged people with and without senile dementia of the Alzheimer type. *J Pathol* 1992; 167: 365-8.
128. Ball MJ. "Limbic" predilection in Alzheimer dementia: is reactivated herpesvirus involved? *Can J Neurol* 1982; 9: 303-6.

129. Ball MJ, Lukiw WJ, Kammermann EM, Hill JM. Intracerebral propagation of  
Alzheimer's disease: strengthening evidence of a herpes virus etiology. *Alzheimers  
Dement* 2013; 9:169-75. doi: 10.1016/j.jalz.2012.07.005.
130. Gibson KL, Wu YC, Barnett Y, Duggan O, Vaughan R, Kondeatis E, et al. B-cell  
diversity decreases in old age and is correlated with poor health status. *Aging Cell*  
2009; 8:18-25.
131. Wozniak MA, Shipley SJ, Combrinck M, Wilcock GK, Itzhaki RF. Productive herpes  
simplex virus in brain of elderly normal subjects and Alzheimer's disease patients. *J  
Med Virol* 2005; 75: 300-6.
132. Lin WR, Graham J, MacGowan SM, Wilcock GK, Itzhaki RF. Alzheimer's disease,  
herpes virus in brain, apolipoprotein E4 and herpes labialis. *Alzheimer's Rep* 1998; 1:  
173-8.
133. Urovesic N, Martins RN. Infection and Alzheimer's disease: the APOE epsilon4  
connection and lipid metabolism. *J Alzheimer's Dis* 2008; 13: 421-35.
134. Lövheim H, Gilthorpe J, Adolfsson R, Nilsson LG, Elgh F. Reactivated herpes  
simplex infection increases the risk of Alzheimer's disease. *Alzheimers Dement* 2014  
pii: S1552-5260(14)02421-2. doi: 10.1016/j.jalz.2014.04.522.
135. Kobayashi N, Nagata T, Shinagawa S, Oka N, Shimada K, Shimizu S, et al. Increase  
in the IgG activity index due to herpes simplex virus type 1 reactivation and its  
relationship with cognitive function in amnesic mild cognitive impairment and  
Alzheimer's disease. *Biochem Biophys Res Commun* 2013; 430: 907-11. doi:  
10.1016/j.bbrc.2012.12.054.
136. Letenneur L, Pérès K, Fleury H, Garrigue I, Barberger-Gateau P, Helmer C, et al.  
Seropositivity to herpes simplex virus antibodies and risk of Alzheimer's disease: a  
population-based cohort study. *PLoS One* 2008; 3: e3637.

- 1015 137. Hill MJ, Zhao Y, Clement C, Neumann DM, Lukiw WJ. HSV-1 infection of human  
1016 brain cells induces miRNA-146a and Alzheimer-type inflammatory signaling.  
1017 Neuroreport 2009; 20: 1500-5. doi: 10.1097/WNR.0b013e3283329c05.
- 1018 138. Lewis RE. Overview of the changing epidemiology of candidemia. Curr Med Res  
1019 Opin 2009; 25:1732-40. doi: 10.1185/03007990902990817.
- 1020 139. Miceli MH, Diaz JA, Lee SA. Emerging opportunistic yeast infections. Lancet Infect  
1021 Dis 2011; 11:142-51. doi: 10.1016/S1473-3099(10)70218-8.
- 1022 140. Song X, Eribe ER, Sun J, Hansen BF, Olsen I. Genetic relatedness of oral yeasts  
1023 within and between patients with marginal periodontitis and subjects with oral health.  
1024 J Periodontal Res 2005; 40: 446-52.
- 1025 141. Kumar J, Sharma R, Sharma M, Prabhavathi V, Paul J, Chowdary CD. Presence of  
1026 *Candida albicans* in root canals of teeth with apical periodontitis and evaluation of  
1027 their possible role in failure of endodontic treatment. J Int Oral Health 2015; 7: 42-5.
- 1028 142. Olsen I. Denture stomatitis. Occurrence and distribution of fungi. Acta Odontol Scand  
1029 1974; 32: 329-33.
- 1030 143. Alonso R, Pisa D, Marina AI, Morato E, Rábano A, Carrasco L. Fungal infections in  
1031 patients with Alzheimer's disease. J Alzheimer's Dis 2014; 41: 301-11. doi:  
1032 10.3233/JAD-132681.
- 1033 144. Alonso R, Pisa D, Rábano A, Carrasco L. Alzheimer's disease and disseminated  
1034 mycoses. Eur J Clin Microbiol Infect Dis 2014; 33: 1125-32. doi: 10.1007/s10096-  
1035 013-2045-z.
- 1036 145. Castellani RJ, Perry G, Smith MA. The role of novel chitin-like polysaccharides in  
1037 Alzheimer disease. Neurotox Res 2007; 12: 269-74.
- 1038 146. Watabe-Rudolph M, Song Z, Lausser L, Schnack C, Begus-Nahrman Y, Scheithauer  
1039 MO, et al. Chitinase enzyme activity in CSF is a powerful biomarker of Alzheimer

- disease. *Neurology* 2012; 78: 569-77. doi: 10.1212/WNL.0b013e318247caa1.
147. Pisa D, Alonso R, Juarranz A, Rábano A, Carrasco L. Direct visualization of fungal infection in brains from patients with Alzheimer's disease. *J Alzheimer's Dis* 2015; 43: 613-24. doi: 10.3233/JAD-141386.
148. Ala TA, Doss RC, Sullivan CJ. Reversible dementia: a case of cryptococcal meningitis masquerading as Alzheimer's disease. *J Alzheimer's Dis* 2004; 6: 503-8.
149. Hoffmann M, Muniz J, Carroll E, De Villasante J. Cryptococcal meningitis misdiagnosed as Alzheimer's disease: complete neurological and cognitive recovery with treatment. *J Alzheimer's Dis* 2009; 16: 517-20. doi: 10.3233/JAD-2009-0985.
150. Singhrao SK, Harding A, Simmons T, Robinson S, Kesavalu L. Crean StJ. Oral inflammation, tooth loss, risk factors, and association with progression of Alzheimer's disease. *J Alzheimer's Disease* 2014; 42: 723-37.
151. Olsen I. Update on bacteraemia related to dental procedures. *Transfus Apher Sci* 2008; 39: 173-8. doi: 10.1016/j.transci.2008.06.008.
152. Tomas I, Diz P, Tobias A, Scully C, Donos N. Periodontal health status and bacteraemia from daily oral activities: systematic review/meta-analysis. *J Clin Periodontol* 2012; 39: 213-28.
153. Navarathna DH, Munasinghe J, Lizak MJ, Nayak D, McGavern DB, Roberts DD. MRI confirms loss of blood-brain barrier integrity in a mouse model of disseminated candidiasis. *NMR Biomed* 2013; 26: 1125-34. doi: 10.1002/nbm.2926.
154. Holmes C, Cotterell D. Role of infection in the pathogenesis of Alzheimer's disease. *CNS Drugs* 2009; 23: 993-1002. doi: 10.2165/11310910-000000000-00000.
155. Bell RD, Winkler EA, Singh I, Sagare AP, Deane R, Wu Z, et al. Apolipoprotein E controls cerebrovascular integrity via cyclophilin A. *Nature* 2012; 485: 512-6. doi:

- 1065 10.1038/nature11087.
- 1066 156. Ujiie M, Dickstein DL, Carlow DA, Jefferies WA. Blood-brain barrier permeability  
1067 precedes senile plaque formation in an Alzheimer disease model. *Microcirculation*  
1068 2003; 10: 463-70.
- 1069 157. Fry M, Ferguson AV. The sensory circumventricular organs: brain targets for  
1070 circulating signals controlling ingestive behavior. *Physiol Behav* 2007; 91: 413-23.
- 1071 158. Mann DM, Tucker CM, Yates PO. Alzheimer's disease: an olfactory connection?  
1072 *Mech Ageing Dev* 1988; 42: 1-15.
- 1073 159. Danielyan L, Schäfer R, von Ameln-Mayerhofer A, Buadze M, Geisler J, Klopfer T,  
1074 al. Intranasal delivery of cells to the brain. *Eur J Cell Biol* 2009; 88: 315-24. doi:  
1075 10.1016/j.ejcb.2009.02.001.
- 1076 160. Johnson NJ, Hanson LR, Frey WH. Trigeminal pathways deliver a low molecular  
1077 weight drug from the nose to the brain and orofacial structures. *Mol Pharm* 2010; 7:  
1078 884-93. doi: 10.1021/mp100029t.
- 1079 161. Leung JY, Chapman JA, Harris JA, Hale D, Chung RS, West AK, et al. Olfactory  
1080 ensheathing cells are attracted to, and can endocytose, bacteria. *Cell Moll Life Sci*  
1081 2008; 65: 2732-9. doi: 10.1007/s00018-008-8184-1.
- 1082 162. Kovács T, Cairns NJ, Lantos PL. beta-amyloid deposition and neurofibrillary tangle  
1083 formation in the olfactory bulb in ageing and Alzheimer's disease. *Neuropathol Appl*  
1084 *Neurobiol* 1999; 25: 481-91.
- 1085 163. Wesson DW, Levy E, Nixon RA, Wilson DA. Olfactory dysfunction correlates with  
1086 amyloid-beta burden in an Alzheimer's disease mouse model. *J Neurosci* 2010; 30:  
1087 505-14. doi: 10.1523/JNEUROSCI.4622-09.2010.
- 1088 164. Goate A, Chartier-Harlin MC, Mullan M, Brown J, Crawford F, Fidani L, et al.  
1089 Segregation of a missense mutation in the amyloid precursor protein gene with

- 1090 familial Alzheimer's disease. *Nature* 1991; 349: 704-6.
- 1091 165. Levy-Lahad E, Wasco W, Poorkaj P, Romano DM, Oshima J, Pettingell WH, Jondro  
 1092 PD, et al. Candidate gene for the chromosome 1 familial Alzheimer's disease locus.  
 1093 *Science* 1995; 269: 973-7.
- 1094 166. Rogaev EI, Sherrington R, Rogaeva EA, Levesque G, Ikeda M, Liang Y, et al.  
 1095 Familial Alzheimer's disease in kindreds with missense mutations in a gene on  
 1096 chromosome 1 related to the Alzheimer's disease type 3 gene. *Nature* 1995; 376: 775-  
 1097 8.
- 1098 167. Nicoll JA, Mrak RE, Graham DI, Stewart J, Wilcock G, MacGowan S, et al.  
 1099 Association of interleukin-1 gene polymorphisms with Alzheimer's disease. *Ann Neurol*  
 1100 2000; 47: 365-8.
- 1101 168. McGeer PL, McGeer EG. History of innate immunity in neurodegenerative disorders.  
 1102 *Front Pharmacol* 2011; 2:77. doi: 10.3389/fphar.2011.00077.
- 1103 169. Kornman KS, Crane A, Wang HY, di Giovine FS, Newman MG, Pirk FW, et al. The  
 1104 interleukin-1 genotype as a severity factor in adult periodontal disease. *J Clin*  
 1105 *Periodontol* 1997; 24: 72-7.
- 1106 170. Galbraith GM, Hendley TM, Sanders JJ, Palesch Y, Pandey JP. Polymorphic cytokine  
 1107 genotypes as markers of disease severity in adult periodontitis. *J Clin Periodontol*  
 1108 1999; 26: 705-9.
- 1109 171. Shao MY, Huang P, Cheng R, Hu T. Interleukin-6 polymorphisms modify the risk of  
 1110 periodontitis: a systematic review and meta-analysis. *J Zhejiang Univ Sci B* 2009; 10:  
 1111 920-7. doi: 10.1631/jzus.B0920279.
- 1112 172. Di Bona D, Candore G, Franceschi C, Licastro F, Colonna-Romano G, Cammà C, et  
 1113 al.  
 1114 Systematic review by meta-analyses on the possible role of TNF-alpha polymorphisms



- in association with Alzheimer's disease. *Brain Res Rev* 2009; 61: 60-8. doi: 10.1016/j.brainresrev.2009.05.001.
173. Yang W, Jia Y, Wu H. Four tumor necrosis factor alpha genes polymorphisms and periodontitis risk in a Chinese population. *Hum Immunol* 2013; 74: 1684-7. doi: 10.1016/j.humimm.2013.08.009.
174. Lambert JC, Ibrahim-Verbaas CA, Harold D, Naj AC, Sims R, Bellenguez C, et al. Meta-analysis of 74,046 individuals identifies 11 new susceptibility loci for Alzheimer's disease. *Nat Genet* 2013; 45: 1452-8. doi: 10.1038/ng.2802.
175. Yuan H, Xia Q, Ge P, Wu S. Genetic polymorphism of interleukin 1 $\beta$ -511C/T and susceptibility to sporadic Alzheimer's disease: a meta-analysis. *Mol Biol Rep* 2013; 40: 1827-34. doi: 10.1007/s11033-012-2237-0.
176. Di Bona D, Plaia A, Vasto S, Cavallone L, Lescai F, Franceschi C, et al. Association between the interleukin-1beta polymorphisms and Alzheimer's disease: a systematic review and meta-analysis. *Brain Res Rev* 2008; 59: 155-63. doi: 10.1016/j.brainresrev.2008.07.003
177. Zhu XC, Tan L, Jiang T, Tan MS, Zhang W, Yu JT. Association of IL-12A and IL-12B polymorphisms with Alzheimer's disease susceptibility in a Han Chinese population. *J Neuroimmunol* 2014; 274: 180-4. doi: 10.1016/j.jneuroim.2014.06.026.
178. Payão SL, Gonçalves GM, de Labio RW, Horiguchi L, Mizumoto I, Rasmussen LT, et al. Association of interleukin 1 $\beta$  polymorphisms and haplotypes with Alzheimer's disease. *J Neuroimmunol* 2012; 247: 59-62. doi: 10.1016/j.jneuroim.2012.03.012.
179. Wang B, Zhou S, Yang Z, Xie YC, Wang J, Zhang P, et al. Genetic analysis of tumor necrosis factor-alpha (TNF-alpha) G-308A and Saitohin Q7R polymorphisms with Alzheimer's disease. *J Neurol Sci* 2008; 270: 148-51. doi: 10.1016/j.jns.2008.02.021.
180. Lio D, Annoni G, Licastro F, Crivello A, Forte GI, Scola L, et al. Tumor necrosis

- factor-alpha -308A/G polymorphism is associated with age at onset of Alzheimer's disease. *Mech Ageing Dev* 2006; 127: 567-71.
181. Kornman KS. Interleukin 1 genetics, inflammatory mechanisms, and nutrigenetic opportunities to modulate diseases of aging. *Am J Clin Nutr* 2006; 83: 475S-83S.
182. Sando SB, Melquist S, Cannon A, Hutton ML, Sletvold O, Saltvedt I, et al. APOE epsilon 4 lowers age onset and is a high risk factor for Alzheimer's disease: a case control study from central Norway. *BMC Neurol* 2008; 8, 9. doi: 10.1186/1471-2377-8-9.
183. Desikan RS, Schork AJ, Wang Y, Thompson WK, Dehghan A, Ridker PM, et al. Polygenic overlap between C-Reactive Protein, plasma lipids and Alzheimer's Disease. *Circulation* 2015 pii: doi: 10.1161/CIRCULATIONAHA.115.015489.
184. Engelborghs S, Gilles C, Ivanoiu A, Vandewoude M. Rationale and clinical data supporting nutritional intervention in Alzheimer's disease. *Acta Clin Belg* 2014; 69: 17-24. doi: 10.1179/0001551213Z.0000000006.
185. Cerajewska TL, Davies M, West NX. Periodontitis: a potential risk factor for Alzheimer's disease. *Brit Dent J* 2015; 218: 29-34.
186. Cicciù M, Maticena G, Signorino F, Brugaletta A, Cicciù A, Bramanti E. Relationship between oral health and its impact on the quality life of Alzheimer's disease patients: a supportive care trial. *Int J Clin Exp Med* 2013; 6: 766-72.
187. Stewart R, Sabbah W, Tsakos G, D'Aiuto F, Watt RG. Oral health and cognitive function in the Third National Health and Nutrition Examination Survey (NHANES III). *Psychosom Med* 2008; 70: 936-41. doi: 10.1097/PSY.0b013e3181870aec.
188. Stein PS, Kryscio RJ, Desrosiers M, Donegan SJ, Gibbs MB. Tooth loss, apolipoprotein E, and decline in delayed word recall. *J Dent Res* 2010; 89: 473-7. doi:

- 10.1177/0022034509357881.
189. Gatz M, Mortimer JA, Fratiglioni L, Johansson B, Berg S, Reynolds CA, et al. Potentially modifiable risk factors for dementia in identical twins. *Alzheimers Dement* 2006; 2: 110-7. doi: 10.1016/j.jalz.2006.01.002.
190. Kaye EK, Valencia A, Baba N, Spiro A 3rd, Dietrich T, Garcia RI. Tooth loss and periodontal disease predict poor cognitive function in older men. *J Am Geriatr Soc* 2010; 58: 713-8. doi: 10.1111/j.1532-5415.2010.02788.x.
191. de Souza Rolim T, Fabri GM, Nitrini R, Anghinah R, Teixeira MJ, de Siqueira JT, Cestari JA, de Siqueira SR. Oral infections and orofacial pain in Alzheimer's disease: a case-control study. *J Alzheimers Dis* 2014; 38: 823-9. doi: 10.3233/JAD-131283.
192. Corrada MM, Paganini-Hill A, Berlau DJ, Kawas CH. Apolipoprotein E genotype, dementia, and mortality in the oldest old: the 90+ Study. *Alzheimers Dement* 2013; 9:12-8.
193. Loeb MB, Molloy DW, Smieja M, Standish T, Goldsmith CH, Mahony J, et al. A randomized, controlled trial of doxycycline and rifampin for patients with Alzheimer's disease. *J Am Geriatr Soc* 2004; 52: 381-7.
194. Ferretti MT, Allard S, Partridge V, Ducatzenzeiler A, Cuello AC. Minocycline corrects early, pre-plaque neuroinflammation and inhibits BACE-1 in a transgenic model of Alzheimer's disease-like amyloid pathology. *J Neuroinflammation* 2012; 9: 62. doi: 10.1186/1742-2094-9-62.
195. in't Veld BA, Ruitenberg A, Hofman A, Stricker BH, Breteler MM. drugs and incidence of dementia: the Rotterdam Study. *Neurobiol Aging* 2001; 22: 407-12.
196. Stewart WF, Kawas C, Corrada M, Metter EJ. Risk of Alzheimer's disease and duration of NSAID use. *Neurology* 1997; 48: 626-32.

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- 1190 [196-197.](#) McGeer PL, McGeer EG. Anti-inflammatory drugs in the fight against  
 1191 disease. *Ann N Y Acad Sci* 1996; 777: 213-20.
- 1192 [197-198.](#) Itzhaki R, Wozniak MA. Could antivirals be used to treat Alzheimer's disease.  
 1193 Future  
 1194 Microbiol 2012; 7: 307-9.
- 1195 [198-199.](#) Lin WR, Jennings R, Smith TL, Wozniak MA, Itzhaki RF. Vaccination  
 1196 prevents latent  
 1197 HSV1 infection of mouse brain. *Neurobiol Aging* 2001; 22: 699-703.
- 1198 [199-200.](#) Mori I. "Spontaneous molecular reactivation" of herpes simplex virus type 1 in  
 1199 the  
 1200 brain as a pathogenic mechanism of Alzheimer's disease. *Correspondence/Med*  
 1201 *Hypotheses* 2011; 77: 462.
- 1202 [200-201.](#) Wozniak MA, Itzhaki RF. Intravenous immunoglobulin reduces  $\beta$  amyloid and  
 1203 abnormal tau formation caused by herpes simplex virus type 1. *J Neuroimmunol* 2013;  
 1204 257: 7-12. doi: 10.1016/j.jneuroim.2013.01.005.
- 1205 [201-202.](#) Jalkute CB, Sonawane KD. Evaluation of a possible role of *Stigmatella*  
 1206 *aurantiaca*  
 1207 ACE in A $\beta$  peptide degradation: a molecular modeling approach. *J Mol Microbiol*  
 1208 *Biotechnol* 2015; 25: 26-36. doi: 10.1159/000370114.
- 1209 [202-203.](#) Chiarini A, Gardenal E, Whitfield JF, Chakravarthy B, Armato U, DaI Pra I.  
 1210 Preventing the spread of Alzheimer's disease neuropathology: a role for calcilytics?  
 1211 *Curr Pharm Biotechnol* 2015 May 5. [Epub ahead of print].
- 1212 [203-204.](#) Hochgräfe K, Sydow A, Matenia D, Cadinu D, Könen S, Petrova O, et al.  
 1213 methylene blue treatment preserves cognition in mice expressing full-length pro-  
 1214 aggregant human Tau. *Acta Neuropathol Commun* 2015; 3: 25. doi: 10.1186/s40478-

1215 015-0204-4.

1216 [204-205.](#) Richard T, Pawlus AD, Iglésias ML, Pedrot E, Waffo-Teguo P, Mérillon JM, et  
 1217 al.

1218 Neuroprotective properties of resveratrol and derivatives. Ann NY Acad Sci 2011;  
 1219 1215: 103-8. doi: 10.1111/j.1749-6632.2010.05865.x.

1220 [205-206.](#) Regitz C, Fitzenberger E, Mahn FL, Dußling LM, Wenzel U. Resveratrol  
 1221 amyloid-beta (A $\beta$ 1-42)-induced paralysis through targeting proteostasis in an  
 1222 Alzheimer model of *Caenorhabditis elegans*. Eur J Nutr 2015 Apr 8. [Epub ahead of  
 1223 print].

1224 [206-207.](#) Håheim LL, Olsen I, Rønningen KS. Oral infection, regular alcohol drinking  
 1225 pattern,  
 1226 and myocardial infarction. Med Hypotheses 2012; 79: 725-30. doi:  
 1227 10.1016/j.mehy.2012.08.010.

1228 [207-208.](#) Song Y, Kim HD, Lee MK, Kim MK, Kang SN, Ko YG, et al. Protective  
 1229 effect of  
 1230 centipede grass against A $\beta$  oligomerization and A $\beta$ -mediated cell death in PC12 cells.  
 1231 Pharm Biol 2015; 8: 1-7.

1232 [208-209.](#) Kontsekova E, Zilka N, Kovacech B, Novak P, Novak M. First-in-man tau  
 1233 vaccine  
 1234 targeting structural determinants essential for pathological tau-tau interaction reduces  
 1235 tau oligomerisation and neurofibrillary degeneration in an Alzheimer's disease model.  
 1236 Alzheimers Res Ther 2014; 1; 6:44. doi: 10.1186/alzrt278.HillH pylori.

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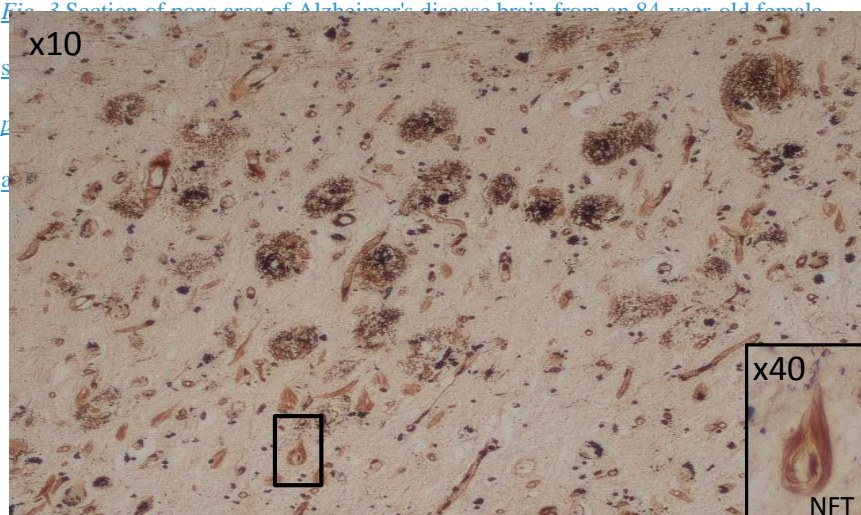
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*Fig. 1.* The pathological hallmarks of AD, numerous extracellular amyloid-A $\beta$  plaques and intra-neuronal neurofibrillary tangles (NFTs). Although there are several NFTs, only one is picked out in boxes at x 10 and x 40 objective lens magnification.

*Fig. 2.* Immunofluorescence labelling (green dots) of hippocampal CA neurons opsonised by iC3b following monoinfection with *P. gingivalis* at 24 weeks of *APOE* gene knockout (*ApoE*<sup>-/-</sup>) mice. This is indirect evidence of an oral infection having affected the host's brain.

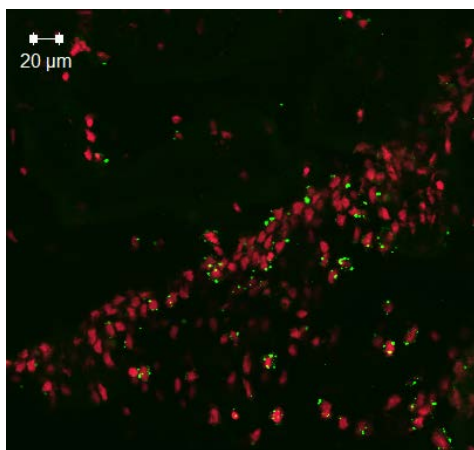
*Fig. 3.* Photo of a Sabouraud agar model made from the upper denture of an old patient with denture stomatitis and heavy accumulations of denture plaque on the fitting surface. *Candida* species are growing profusely.

*Fig. 4* Section of non-neuron of Alzheimer's disease brain from an 84-year-old female



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*Fig. 1.*

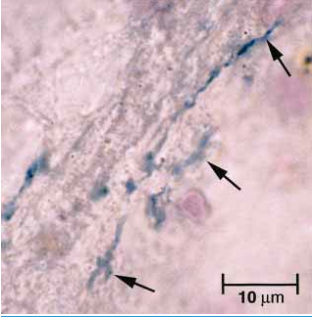


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*Fig. 2.*

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*Fig. 3.*